Remarks:

In the Office Action dated November 7, 2004, claims 53, 57-58 and 60-103, in the above-identified U.S. patent application were rejected. Reconsideration of the rejections is respectfully requested in view of the above amendments, the remarks in applicant's January 19, 2005 response and the following remarks. Claims 53, 57, 60-63 and 65-103, remain in this application, claims 1-52, 54-56,58 and 64 have been canceled. The limitations from claim 58 have been incorporated into claims 53, 86 and 94. Support for new claim 104 can be found on page 24, lines 11-35 and page 36, lines10-29, of the present specification.

The present claims recite a mixture of hybridisation probes which includes a probe which is specific for the wild type. As discussed on page 22 of the specification, the presence of the wild type sequence increases the sensitivity of the mutation-specific probe (see also Example 2, p. 36). The wild type probe and the mutation probe compete for binding to the sequences of the microorganism. Perfectly matching sequences are favored because the perfectly matching hybridisation complexes have the largest dissociation temperatures. Thus, since hybridisation of the mutation probe alone leads to a certain degree of cross-hybridisation with the wild type sequence, the presence of a wild type probe saturates the wild type sequences in the microorganisms and thereby reduces cross-hybridisation, leading to improved specificity.

In mutated sequences of the present invention, an A can be replaced by G or C which leads to an increased dissociation temperature of about 2 °C

(see Formula on page 48, lines 6-10), and the mutation probe hybridises under even more stringent conditions with its exactly complementary sequence compared with the wild type sequence. The length of the probes in the examples in the present application is 17 nucleotides and thus the mutated sequence has an increased CG content of about 5% compared with the wild type. By increasing the CO content of the mutation probe (e.g. A to C and A to G mutations), the amount of cross-hybridisation of the mutation probe can be further reduced. When a wild type probe and a mutation probe are applied at the same time, the following situation occurs for the probes of the present invention having a length of 17 nucleotides:

	Microorganism	Probe	Hybridisation complex	Dissociation temp. of Hybridisation complex
1	WT	WT	WT-WT	X°C
2		Mutation (labeled)	WT-Mutation	<X°C
3	Mutation (A->C or A->G)	WT	Mutation-WT	<X°C
4		Mutation (labeled)	Mutation-Mutation	X+2°C

X is the dissociation temperature of the WT-WT complex. The dissociation temperatures of WT-Mutation (line 2 of the Table) and Mutation-WT (line 3) should be identical.

The table shows that by a A to C or A to G transition, the difference of the dissociation temperatures of the Mutation-WT and Mutation-Mutation complex increases and thereby favors binding of the mutation probe to the mutation

sequence, up to 100%. This effect improves specificity. Thus, the wild type probe may not bind to the mutation sequence.

Applicants point out that none of the cited prior art suggests that macrolide antibiotic resistance can be detected using a mixture of hybridization probes which specifically detect point mutations and which includes a hybridization probe specific for a wild type nucleic acid sequence as in the present invention. The recited mixture of hybridization probes results in increased sensitivity which enables the present invention to detect a sequence difference of only one single base.

Applicants also point out that table 7 on page 41 of the present application provides a sequence alignment of the region surrounding E. coli positions 2057 to 2059 with the sequences of more than 40 microorganism species and strains. rRNAs in different organisms usually have different lengths but the positions corresponding to E. coli positions 2057 to 2059 can easily be determined using such an alignment. The same strategy applies to E. coli positions 2032 and 2611 (i.e. the corresponding sequences in other microorganisms can be determined by alignment).

Applicants respectfully submit that all of claims 53, 57, 59-63 and 65-104 are now in condition for allowance. If it is believed that the application is not in condition for allowance, it is respectfully requested that the undersigned attorney be contacted at the telephone number below.

In the event this paper is not considered to be timely filed, the Applicant respectfully petitions for an appropriate extension of time. Any fee for such an

extension together with any additional fees that may be due with respect to this paper, may be charged to Counsel's Deposit Account No. 02-2135.

Respectfully submitted,

By _____

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